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Chemical compositions of *Casuarina equisetifolia* L., *Eucalyptus toreliana* L. and *Ficus elastica* Roxb. ex Hornem cultivated in Nigeria

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Abstract

Essential oils were obtained by separate hydrodistillation of three different plants cultivated in Nigeria and analysed comprehensively for their constituents by means of gas chromatography (GC) and gas chromatography-mass spectrometry (GC–MS). The leaf essential oil of *Casuarina equisetifolia* L. (Casuarinaceae) comprised mainly of pentadecanal (32.0%) and 1,8-cineole (13.1%), with significant amounts of apiole (7.2%), α-phellandrene (7.0%) and α-terpinene (6.9%), while the fruit oil was dominated by caryophyllene-oxide (11.7%), *trans*-linalool oxide (11.5%), 1,8-cineole (9.7%), α-terpineol (8.8%) and α-pinene (8.5%). On the other hand, 1,8-cineole (39.4%) and α-terpinyl acetate (10.7%) occurred in large quantities in the essential oils of the leaf of *Eucalyptus toreliana* L. (Myrtaceae). The oil also features high levels of sabinene (5.9%), caryophyllene-oxide (4.7%) and α-pinene (4.2%). The main compounds identified in the leaf oil of *Ficus elastica* Roxb. ex Hornem. (Moraceae) were 6,10,14-trimethyl-2-pentadecanone (25.9%), geranyl acetone (9.9%), heneicosene (8.4%) and 1,8-cineole (8.2%).

Keywords: 1,8-Cineole; 6,10,14-Trimethyl-2-pentadecanone; Caryophyllene-oxide; *Casuarina equisetifolia*; Essential oil; *Eucalyptus toreliana*; *Ficus elastica*; Pentadecanal; *trans*-Linalool oxide; α-Terpinyl acetate

1. Introduction

Casuarina is a genus of 17 species in the family Casuarinaceae, native to Australasia, southeastern Asia, and islands of the western Pacific Ocean. *Casuarina equisetifolia* L. is a widespread seashore tree known as Common Ironwood, Beefwood, Bull-oak, and Whistling-pine and is often planted as a windbreak. This species is not a pine at all, but superficially resembles a conifer (Pinyopusarerk and House, 1993). The plant is a source of biologically active compounds such as catechin, ellagic acid, gallic acid, quercetin and lupeol, which are antioxidants (Aher et al., 2009), coumaroyl triterpenes (Takahashi et al., 1999) and *d*-gallocatechin (casuarin) (Nash et al., 1994). The plant is also known to store tannins (Li-Hua et al., 2009) and proline (Tani and Sasakawa, 2003) as well as being a nitrogen fixing plant (Li-Hua et al., 2009). *C. equisetifolia* also displays antimicrobial properties (Parekh et al., 2005).

Eucalyptus (Myrtaceae) is a large genus of trees and shrubs, which originates mainly from Australia. *Eucalyptus torreliana* L., is one of the known 500 species of *Eucalyptus* that produces terpenoids. The leaves of the *Eucalyptus* species have medicinal and flavouring properties. The essential oil-bearing *Eucalyptus* plants rank high both in quantity and frequency among the plants that are widely used all over the world (Ogunwande, 2001). Reports on the chemical constituents of some *Eucalyptus* species cultivated in Nigeria have been published (Ogunwande et al., 2003; Ogunwande et al., 2005; Jimoh et al., 2005).

Ficus elastica Roxb. ex Hornem, (also known as the rubber tree) is a common house plant, as it can grow in moderately

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luminous environments. As with other members of the genus *Ficus*, the flowers require a particular species of fig wasp to pollinate it in a co-evolved relationship. Rubber plants are not known to produce highly colourful or fragrant flowers to attract other pollinators (Busari, 2001). The edible fruit is a small yellow-green oval fig, one centimetre long and barely edible.

2. Materials and methods

2.1. Plant materials

Mature leaves and fruits of *C. equisetifolia* were collected from trees growing in front of Okunuga Hall, Faculty of Law, Lagos State University, Ojo, in March 2009. The leaves of *E. torreliana* were harvested from trees growing at Eleyele, a suburb of Ibadan, Oyo State, Nigeria, in April 2009, while those of *F. elastica* were obtained from the Botanical Garden, University of Lagos, Nigeria, in March 2009. The samples of *C. equisetifolia* and *F. elastica* were identified by Messrs. Ugbuoga and Shosanya at the Herbarium Headquarters, Forestry Research Institute of Nigeria (FRIN), Ibadan, where voucher specimens (FHI 107885 for *C. equisetifolia* and FHI 102907 for *F. elastica*), were deposited. *E. torreliana* was identified by Mr. Ogunduyile of the Herbarium, Department of Botany and Microbiology, University of Ibadan, Nigeria, where voucher specimen (UIH 222244) was kept for future reference.

2.2. Isolation of the volatile oils

Aliquots of air-dried and pulverised samples were hydrodistilled in an all glass Clevenger-type apparatus for 3 h according to established procedure (British Pharmacopoeia, 1990). Aliquots of leaves (1.0 kg) and fruits (0.40 kg) of *C. equisetifolia* afforded pale yellow oils at yields of 0.09 and 0.10% (v/w), respectively. In addition, 0.45 and 0.95 kg of *E. torreliana* and *F. elastica* afforded colourless and pale yellow oils, respectively, at yields of 0.34 and 0.11% (v/w).

2.3. Gas chromatography (GC) and gas chromatography–mass spectrometry (GC–MS)

GC analysis was accomplished with an HP-5890 Series II instrument equipped with HP-Wax and HP-5 capillary columns (both 30 m × 0.25 mm, 0.25 μm film thickness), with the following temperature programme: 60 °C for 10 min, rising at 5 °C/min to 220 °C. Both injector and detector temperatures were maintained at 250 °C; carrier gas nitrogen (2 mL/min); detector, FID; split ratio 1:30. The volume injected was 0.5 μL. The identification of the components was performed by comparison of their retention times with those of pure authentic samples and by means of their linear retention indices (LRI on HP-5 column) relative to the series of *n*-hydrocarbons. The relative proportions of the oil constituents were percentages obtained (% area) by FID peak-area normalisation without the use of response factor.

Gas chromatography–electron ionisation mass spectrometry analysis was performed with a Varian CP-3800 gas-chromatograph equipped with a HP-5 capillary column (30 m × 0.25 mm;

Table 1
Constituents of the leaves and fruits of *Casuarina equisetifolia*.

Constituents	LRI	Percentage	
		Leaves	Fruits
(E)-2-hexenal	854	0.1	–
heptanal	899	0.5	–
tricyclene	926	tr	–
α-thujene	931	0.5	–
α-pinene	939	1.9	8.5
camphene	953	1.3	–
benzaldehyde	961	tr	3.4
sabinene	976	2.8	–
β-pinene	980	0.8	–
6-methyl-5-hepten-2-one	985	tr	–
myrcene	991	1.6	–
δ-2-carene	1001	tr	–
α -phellandrene	1007	7.0	–
δ-3-carene	1012	tr	–
δ-terpinene	1020	6.9	–
p-cymene	1028	3.7	–
limonene	1033	1.9	–
1,8-cineole	1035	13.1	9.7
(Z)-β-ocimene	1042	tr	–
(E)-β-ocimene	1052	tr	–
α-terpinene	1063	0.9	–
cis-sabinene hydrate	1071	tr	–
octanol	1072	tr	–
terpinolene	1090	tr	–
trans-linalool oxide (furanoid)	1095	–	11.5
linalool	1100	0.8	–
nonanal	1104	1.6	–
α-campholenal	1131	–	5.2
4-terpineol	1179	tr	4.6
α-terpineol	1191	tr	8.8
safranal	1202	tr	–
decanal	1206	tr	–
cis-ascaridole	1237	0.7	–
neral	1240	tr	–
geranial	1272	tr	–
2-undecanone	1293	tr	–
tridecane	1300	tr	–
α-terpinyl acetate	1355	–	6.2
eugenol	1358	tr	–
α-copaene	1377	tr	–
β-cubebene	1391	tr	–
β-elemene	1393	tr	–
tetradecane	1400	0.4	–
methyl eugenol	1403	tr	–
dodecanal	1409	0.6	–
β-caryophyllene	1420	0.5	–
(E)-α-ionone	1430	tr	–
1-methoxynapthalene	1446	0.3	–
geranyl acetone	1455	0.8	–
alloaromadendrene	1461	tr	–
γ-muurolene	1477	tr	–
germacrene D	1482	tr	–
(E)-β-ionone	1486	0.8	–
cis-eudesma-6,11-diene	1490	0.2	–
bicyclogermacrene	1495	0.2	–
α-selinene	1496	tr	–
pentadecane	1500	tr	–
β-bisabolene	1509	0.2	–
tridecanal	1510	0.5	–
myristicin	1520	tr	–
β-sesquiphellandrene	1525	1.4	–
germacrene B	1557	0.2	–

Table 1 (*continued*)

Constituents	LRI	Percentage	
		Leaves	Fruits
trans-nerolidol	1566	0.1	–
spathulenol	1578	–	4.4
caryophyllene-oxide	1583	–	11.7
guaiol	1595	–	6.4
hexadecane	1600	tr	–
tetradecanal	1613	4.7	–
τ-cadinol	1642	tr	–
α-cadinol	1655	tr	–
cadalene	1676	tr	–
apiole	1680	7.2	–
heptadecane	1700	tr	–
pentadecanal	1719	32.0	–
octadecane	1800	0.2	–
hexahydrofarnesylacetone	1848	0.9	–
Total identified		97.3%	80.4%

Linear retention indices on HP-5 capillary column; tr, trace amount<0.1%.
–, not identified.

film thickness 0.25 μm) and a Varian Saturn 2000 ion trap mass detector. Analytical conditions: injector and transfer line temperature 220 °C and 240 °C, respectively; oven temperature programmed from 60 to 240 °C, at 3 °C/min.; carrier gas was helium at a flow rate of 1 mL/min.; injection of 0.2 μL (10% hexane solution); split ratio 1:30. Mass spectra were recorded at 70 eV. The acquisition mass range was 30–300 m/z at a scan rate of 1 scan/s.

Identification of the constituents was based on comparison of the retention times with those of authentic samples, comparing their linear indices relative to the series of *n*-hydrocarbons, and on computer matching against commercially available spectral (Adams, 2005). Further identifications were also made possible by the use of self constructed spectral library built up from pure substances and components of known oils and MS literature data (Davies, 1990; Jennings and Shibamoto, 1980; Massada, 1975). Moreover, the molecular weights of all the identified substances were confirmed by gas chromatography–chemical ionisation mass spectrometry, using methanol as CI ionising gas.

3. Results and discussion

Table 1 lists the compounds identified in the leaf and fruit oils of *C. equisetifolia*. Seventy-six compounds comprising of monoterpene hydrocarbons (29.3%), oxygenated monoterpenoids (16.2%), sesquiterpene hydrocarbons (2.7%), oxygenated derivatives (1.0%), aliphatic (40.6%) and non-terpenoid (7.2%) compounds were observed in the leaf oils. The major compounds were pentadecanal (32.0%) and 1,8-cineole (13.1%). Significant quantities of α-phellandrene (7.0%), apiole (7.2%) and α-terpinene (6.9%) were present. The fruit oil was devoid of sesquiterpene hydrocarbon compounds. The main constituents were caryophyllene-oxide (11.7%), *trans*-linalool oxide (11.5%), 1,8-cineole (9.7%), α-terpineol (8.8%) and α-pinene (8.5%). All the eleven compounds identified in the oil occurred at levels between 3.4 and 11.7%. Both caryophyllene-oxide and *trans*-linalool oxide were absent in the leaf oil. The authors are not aware of any literature report on the constituents of the essential oil of the plant or of any *Casuarina* species and as such the present study may represent the first of its kind.

Table 2
Percentage constituents of *Eucalyptus torreliana*.

Constituents	LRI	Percentage (%)
α-thujene	931	tr
α-pinene	939	4.2
camphene	953	tr
sabinene	976	5.9
β-pinene	980	2.9
myrcene	991	0.8
α-phellandrene	1008	1.0
δ-terpinene	1021	0.8
p-cymene	1029	1.0
limonene	1034	2.0
1,8-cineole	1037	39.4
α-terpinene	1064	0.6
cis-sabinene hydrate	1073	tr
dihydro myrcenol	1074	0.8
terpinolene	1091	tr
linalool	1103	2.2
trans-pinocarveol	1139	tr
borneol	1170	1.3
δ-terpineol	1172	tr
4-terpineol	1181	2.7
α-terpineol	1193	3.9
isobornyl acetate	1290	tr
α-terpinyl acetate	1355	10.7
neryl acetate	1370	tr
β-bourbonene	1385	tr
β-elemene	1394	tr
n-tetradecane	1400	tr
methyl eugenol	1409	1.6
β-caryophyllene	1421	3.4
trans-α-bergamotene	1439	tr
(E)-isoeugenol	1451	tr
α-humulene	1458	0.7
germacrene D	1483	1.6
(E)-β-ionone	1487	tr
(E)-methyl isoeugenol	1495	2.4
α-bulnesene	1506	0.5
trans-nerolidol	1567	tr
spathulenol	1578	0.7
caryophyllene-oxide	1583	4.7
n-hexadecane	1600	tr
eremoligenol	1631	tr
γ-eudesmol	1635	tr
β-eudesmol	1651	tr
pentadecanal	1719	1.4
14-hydroxy-α-muurolene	1780	0.9
Total		99.5%

Linear retention indices on HP-5 capillary column; tr, trace amount<0.1%.

Monoterpenes (85.6%) and sesquiterpenes (12.5%), typical of *Eucalyptus* were the dominant classes of compounds among the forty-five constituents identified from the oil of *E. toreliana*. The aliphatic compounds made up 1.4% of the total oil content. 1,8-Cineole (39.4%) and α-terpinyl acetate (10.7%) constituted sizeable proportion of the oil content (Table 2). Other noteworthy compounds were sabinene (5.9%), caryophyllene-oxide (4.7%) and α-pinene (4.2%). In a previous report (Chalchat et al., 2000), 1,8-cineole, α-pinene, β-pinene and limonene were identified as

Table 3
Compounds of *Ficus elastica*.

Constituents	LRI	Percentage (%)
(*E*)-2-hexenal	854	0.2
2-heptanone	889	tr
α-thujene	931	tr
α-pinene	939	1.2
camphene	953	tr
benzaldehyde	961	0.9
sabinene	976	1.0
β-pinene	980	0.5
6-methyl-5-hepten-2-one	985	tr
δ-2-carene	1001	tr
α-phellandrene	1007	1.4
δ-terpinene	1020	1.1
p-cymene	1028	1.1
limonene	1033	1.7
1,8-cineole	1035	8.2
benzene acetaldehyde	1045	tr
α-terpinene	1063	tr
acetophenone	1067	tr
nonanal	1104	1.1
naphthalene	1181	1.9
safranal	1201	tr
carvenone	1252	tr
(*E*)-2-decenal	1261	tr
2-undecanone	1293	tr
β-caryophyllene	1420	tr
(*E*)-α-ionone	1428	2.4
cis-α-ambrinol	1438	0.2
geranyl acetone	1455	9.9
(*E*)-β-ionone	1486	3.9
butylated hydroxy anisole	1489	tr
2-tridecanone	1496	tr
pentadecane	1500	1.0
α-calacorene	1543	tr
cis-sesquisabinene hydrate	1546	tr
trans-nerolidol	1566	0.8
caryophyllene-oxide	1582	4.2
hexadecane	1600	1.0
humulene epoxide II	1608	tr
tetradecanal	1613	1.4
epoxy-alloaromadendrene	1641	tr
selin-11-en-4α-ol	1654	1.6
neo- intermedeol	1660	tr
5-iso cedranol	1674	1.9
acorenone	1685	1.5
heptadecane	1700	3.3
pentadecanal	1719	6.1
drimenol	1759	1.0
octadecane	1800	1.1
khusinol acetate	1824	tr
6,10,14-trimethyl-2-pentadecanone	1848	25.9
nonadecane	1900	1.1
eicosane	2000	0.9
heneicosene	2096	8.4
heneicosane	2100	0.2
docosane	2200	0.3
Total		98.4%

Linear retention indices on HP-5 capillary column; tr, trace amount<0.1%.

the dominant constituents of the oil of *E. toreliana*. In addition, an earlier report (Jimoh et al., 2005) characterised by only GC–MS revealed the abundance of α-pinene (21.7%), β-pinene (10.3%), β-copaene (16.8%) and 1,8-cineole (32.8%) as constituents identified quantitative importance. The predominance of terpene compounds in the essential oil is typical of *Eucalyptus* species with concurrent variations in the medicinal properties of the oils (Ogunwande, 2001). 1,8-Cineole has been described as the most frequent major compound occurring in *Eucalyptus* oils (Chalchat et al., 2000; Ogunwande, 2001; Ogunwande et al., 2003; Ogunwande et al., 2005).

Aliphatic compounds (52.0%) occurred in highest proportions in the volatile oil of *F. elastica* (Table 3). Monoterpenes (32.6%) and oxygenated sesquiterpenes (11.0%) were also prominent. The main constituents include 6,10,14-trimethyl-2-pentadecanone (25.9%), geranyl acetone (9.9%), heneicosene (8.4%) and 1,8-cineole (8.2%). The other compounds of note were pentadecanal (6.1%), *trans*-nerolidol (4.2%), (*E*)-β-ionone (3.9%) and heptadecane (3.3%). The volatile oil composition of this plant has not been a subject of literature discussion. Previous studies on Nigerian grown *Ficus* species have revealed the abundance of 1,8-cineole (13.8%), (*E*)-phytol (13.7%) and *p*-cymene (11.4%) in *Ficus exasperata* (Sonibare et al., 2006); β-caryophyllene (37.0%), ethyl octanoate (14.9%) and methyl octanoate (8.3%) in *Ficus mucosa* (Ogunwande et al. 2009); acorenone (20.7%) and phytol (16.2%) in *Ficus lutea*, with *Ficus polita* consisting mainly of phytol (23.3%) and 6, 10, 14-trimethyl-2-pentadecanone (15.0%) and *Ficus thonningii* rich in 6, 10, 14-trimethyl-2-pentadecanone (18.8%) and phytol (14.7%) (Ogunwande et al., 2008). Both 6,10,14-trimethyl-2-pentadecanone and phytol have been described as marker components of the oils of Nigerian grown *Ficus* species (Ogunwande et al., 2008). In addition to 6,10,14-trimethyl-2-pentadecanone and 1,8-cineole, other constituents such as acorenone, (*E*)-6, 10-dimethyl-5, 9-undecadien-2-one, ethyl octanoate, methyl octanoate and phytol that are characteristic components of other Nigerian *Ficus* species, are conspicuously absent in *F. elastica*.

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